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Benthic monitoring methods for habitat management of finfish mariculture in Canada

Méthodes de monitorage benthique pour la gestion de l'habitat de pisciculture au Canada

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ABSTRACT

Selected environmental methods chosen to monitor organic enrichment from finfish mariculture sources include those based on macrofaunal taxa and abundance, total sulfide and zinc/copper concentrations in surficial sediments, U/W photo- and video-graphy, and dissolved oxygen in benthic boundary layer seawater. A wide range of parameters and methods to quantify them must be considered because of the variety of habitats on Canada's west and east coasts. We considered methods that are fully accepted within the scientific community and readily available within Canada. The strengths and weaknesses of each method are assessed on the basis of the following criteria: type of sedimentary substrate, scientific defensibility, statistical testability, availability of thresholds and cost effectiveness.

RÉSUMÉ

Les taxons de macrofaune et leur abondance, les concentrations de sulfure total et de zinc/cuivre dans les sédiments superficiels, la photographie et la vidéographie en ultraviolet et la concentration d'oxygène dissous dans la couche limite benthique comptent parmi les méthodes choisies pour surveiller l'enrichissement organique des eaux imputable à la pisciculture. Une vaste gamme de paramètres et de méthodes doivent être considérés pour quantifier ces facteurs en raison de la diversité des habitats retrouvés sur les côtes Est et Ouest du Canada. Nous considérons des méthodes pleinement acceptées par le milieu scientifique et facilement disponibles au Canada. Les atouts et les faiblesses de chacune sont évalués en regard des critères suivants : type de substrat sédimentaire, solidité du point de vue scientifique, testabilité du point de vue statistique, disponibilité de seuils et efficacité par rapport au coût.

INTRODUCTION

One important ecological effect of Canadian intensive finfish mariculture is organic enrichment caused by the waste food and excretory products produced by the fish themselves. The enrichment process which results from such organic wastes occurs both in the water column and in sediments. Consideration of the local environment of the footprint area near the farm or individual fish cage is referred to as near-field. Anything further afield than this is a far-field effect. Summaries of the environmental effects of mariculture in Canada have been presented for the far-field by F&O (2003) and near-field by F&O (2004). These reviews followed the general consensus that the bulk of waste particulates, particularly in net depositional soft sediments, settled in a near-field area. More recent evidence reviewed below suggests that there may be a much wider dispersal of particulates from fish farms than previously thought.

Recent mass balance model calculations in southwestern New Brunswick (SWNB) in locations considered to be net depositional (fine-grained sediments and relatively low current velocities ~5 cm s^{-1}) show that >95% of particulate matter released does not accumulate under pens (Strain and Hargrave 2005). Although some rapidly settling waste particles obviously do accumulate under pens, most of it appears to be transported away from the farm site. A much greater than previously thought dispersal of particulates is explained by the fact that mixtures of feed debris and feces have lower setting velocities (<3 cm s^{-1}) than intact feed pellets. Pellets are also rapidly hydrated and disaggregate into smaller particles with reduced settling rates (Stewart and Grant 2002). The reduced sinking speeds can result in particle transport over distances of 0.1 to 1 km or greater (Cromey et al. 2002; Cromey and Black 2005; Stucchi et al. 2005).

Direct measurements of sedimentation with bottom-mounted sediment traps or traps moored a fixed distance above bottom have confirmed the potential for resuspension, horizontal transport and widespread dispersion of particulate waste products from finfish farms (Brooks 2001; Sutherland 2001a; Cromey et al. 2002; Brooks and Mahnken 2003a; Cromey and Black 2005; Stucchi et al. 2005). Holmer (1991) collected material directly attributable to a finfish aquaculture source at distances up to 1.2 km from a farm site in Danish coastal waters. The extent to which resuspension and lateral transport increase sedimentation at remote locations depends on both physical and sedimentological processes. Processes leading to sediment accumulation, and hence the relative importance of near- and far-field deposition, are highly site-specific and depend on bottom topography, currents, erosion and flocculation processes that affect the residence time of material both in the water column and on the sediment surface (Milligan and Law 2005).

We attempt here to answer two specific questions of interest to DFO Habitat Management concerning environmental effects of finfish mariculture. The questions were formulated at a meeting held in St.Andrews on the 13 – 14th October, 2004 and are:

1. What are the strengths and weaknesses of the existing monitoring methods used to detect and monitor organic enrichment from finfish aquaculture in the marine environment?
2. What threshold effects of environmental change can be used practically for habitat management purposes?

Our aim is to provide answers to these questions from a Canada-wide perspective.

In choosing the short list of environmental methods to include for monitoring organic enrichment effects from mariculture, we have considered only those methods which are

fully developed scientifically for immediate use and which are readily available to DFO Habitat Management personnel throughout Canada. A result of this restriction is that some superior methods are not included here (many can be found in the Further Research section).

Besides practical considerations above the choice of the best method applicable in monitoring of environmental effects of mariculture depends on:

- whether the effect occurs in surficial sediments or the water column;
- what the monitoring goals are (see Table 1);
- determination of the nature of the substrate in sampling the seabed. This is because bottom sampling devices are substrate specific (Fig.1). Methods to do this are considered in the next section.

Choices among competing methods can be made on the basis of scientific and habitat management utility criteria (Holmer et al. 2001). We would expect all candidate methods to have a scientific basis, i.e. be referable to a null and alternate hypothesis (Table 1) and where a statistical test can be used to distinguish between them. Final choices are made by considering whether the method provides suitable threshold limits of environmental change and the lowest comparative cost. The details of sampling design are not considered in this presentation.

DEFINING THE SUBSTRATE TYPES AND BENTHIC HABITATS

Mapping substrates and benthic habitats can be carried out using a combination of continuous, semi-continuous, and discrete mapping techniques. Continuous mapping consists of multibeam surveys which provide a high spatial resolution assessment of substrate types, while discrete mapping consists of spot sampling (grab/core deployments) that provides a discontinuous distribution of seafloor characteristics and benthic habitats. Semi-continuous mapping is an intermediate technique involving both acoustic (QTCView) and video (ROV) surveys along spaced transects within a grid system, or designated transects along organic enrichment gradients. Although semi-continuous and discrete sampling techniques can be used if continuous mapping methods are not available, ideally, the ROV surveys and grab sampling should be used to ground-truth acoustic surveys. Since seafloor substrate type is correlated with the composition of benthic communities, ground-truthed acoustic surveys can often be used to identify benthic habitats (Magorrian et al. 1995; Wildish et al. 1998; Tlusty et al. 2000a; Kostylev et al. 2001).

The extensive spatial coverage obtained by acoustic surveys provides a steering platform for designing smaller-scale, cost-effective monitoring programs that can target substrate types, critical benthic habitats and reference areas. The decision tree presented in Fig. 1 provides a framework for choosing the appropriate monitoring equipment or method according to substrate identified by an acoustic survey (Sutherland 2004). The following description outlines the effectiveness of ground-truthed multibeam surveys in identifying bottom types within three study areas of the DFO Strategic Science Project Environmental Studies for Sustainable Aquaculture (ESSA). Multibeam (EM3000) methods were used to map bathymetry and sediment backscatter intensity in the Broughton Archipelago (Pacific), Bay of Fundy (Maritime), and Bay d'Espoir (Newfoundland) (Hughes-Clarke 2001; Hughes-Clarke et al. 2002; Sutherland 2001b; Tlusty et al. 2000a and 2001; Hargrave 2004; Wildish et al. 2004a). Ground-truthing existed for interpretation of the acoustic results that allowed differentiation of hard and soft bottom areas and maps were prepared for sites of varying size in the three regions. In the Broughton Archipelago,

empirical relationships were derived between the backscatter component of the multibeam survey and sediment grain size fractions along with total sediment sulfides, confirming that the approach could be used to identify substrate types as well as zones of organic enrichment, respectively.

Semi-continuous survey methods such as QTC (a bottom type classification system) surveys were also completed in the Bay d'Espoir and at one site in the Broughton Archipelago. Data from some of these studies have been analyzed to show that acoustic information can differentiate erosional and depositional bottom types and identify areas of gas-filled, fine-grained sediments (Tlusty et al. 2000a; Wildish et al. 2004a). In addition, overlapping multibeam and QTCView surveys in the Broughton Archipelago were both capable of identifying similarly distinct regions of seafloor characterized by rock outcrops, gel-mud depositional fields and impact zones. The RoxAnn™ acoustic bottom discrimination system has also been evaluated to determine its application in identifying benthic impacts due to high sedimentation rates around finfish farm sites (MacDougall and Black 1999), with inconclusive results because the site was erosional.

The availability of EM3000 data in Newfoundland (NF), southwest New Brunswick (SWNB) and British Columbia (BC) in areas of mariculture development allows determination of the relative proportion of soft and hard bottom in these specific areas. The problem is that spatial coverage in each region is incomplete and may not include locations where industry expansion may occur. Equipment to obtain the acoustic information may not be widely available and special expertise is required for data interpretation. Although the information is needed immediately to assist Habitat Management in making decisions regarding approvals for new farm sites, obtaining multibeam data from new locations will take time. Collaboration between the Canadian Hydrographic Service (CHS), Natural Resources Canada (NRCan) and DFO is required to do this. Priority areas for obtaining new acoustic data in specific coastal locations are not always the same between different government departments.

Although both continuous and semi-continuous types of acoustic surveys are necessary to achieve anything other than local maps of benthic habitat, these state-of-the-art technologies may not be readily available to the industry when required and be cost-effective. Since an efficient monitoring program requires the implementation of substrate-specific monitoring tools (Fig. 1), commercially available equipment such as bottom video, photographic surveys and sediment profile imaging (SPI) can provide alternatives to acoustic methods for bottom type/habitat classification and overall assessment of benthic organic enrichment (O'Connor et al. 1989; Valente et al. 1992; Nilsson and Rosenberg 1997, 2000; Wildish et al. 2003; Hargrave et al. 2004). The use of divers in shallow water or mounting equipment on towed bodies or underwater remote vehicles (ROVs) allows observations along survey lines but does not provide the full spatial resolution captured in an EM3000 survey. In the absence of data from this ideal survey method, point or transect sampling using bottom video, still images or SPI are currently the only practical alternatives for determining the proportion of hard and soft substrates in any given area. This pre-survey step in the monitoring decision tree (Fig. 1) is crucial for making decisions about the type of benthic monitoring methods to be used and the provision of baseline data regarding existing benthic habitats.

The concept of scale as an important determinant of ecological pattern has arisen in parallel with the emergence of landscape ecology as a discipline (Schneider 2001). For a recent discussion of the importance of scale and temporal and spatial heterogeneity for

benthic ecology see Raffaelli et al. (2003) and Solan et al. (2003). Two important concepts with respect to organic enrichment from mariculture are pertinent. Ecological processes operate within a hierarchy of scales (Weins 1989; Parry et al. 2003; Brind'Amour et al. 2005), and small scale studies or patterns do not necessarily scale up to larger areas. Thus the question of interest must be matched to the relevant scale, and the methodologies employed must be appropriate to measurement at that scale. If question and scale do not match then we risk extrapolating beyond the limits of the patterns or relationships uncovered. For example, in the Weddell Sea disturbance in the form of iceberg scours significantly reduced species diversity at the local (1-100 m) scale while enhancing it at the regional (1 to 100 km) scale (Gutt and Piepenburg 2003). Studies of landscape processes and patterns have also demonstrated that the grain and extent of the study as well as the classification scheme can induce bias or inaccuracies if the sample and analysis scale are improperly matched to topographic complexity and habitat heterogeneity (Weins 1989; Fuhlendorf and Smeins 1996). Coarse grained data will bias the estimation of small patch sizes, while inappropriate calculation units for data averaging can also induce error in the classification (Weins 1989; Fuhlendorf and Smeins 1996; O'Neill et al. 1996). O'Neill et al. (1996) recommend that the grain of the data be 2-5X smaller than the smallest feature of interest. In consequence, very fine grained information is required to adequately map small scale complexity. Thus topographic relief and substrate heterogeneity are important considerations in the design of seascape, sampling programs with which to survey organic enrichment impacts.

NEAR-FIELD ENVIRONMENTAL HABITAT MONITORING METHODS

Five monitoring methods are available to monitor organic enrichment effects from mariculture in the diverse environmental conditions present within Canada.

Benthic Macrofaunal Species and Abundance in Soft Sediments

Benthic macrofauna are pragmatically defined as those animals which are retained on a sieve of 0.5 to 1.0 mm mesh following sediment sieving. Species included are taxonomically diverse and usually belong to the invertebrate phyla. Their life forms include *infauna*, which live in burrows within the sediment, *epifauna*, which are either attached or free-living at the sediment-water interface, and *tube-builders*, which construct tubular structures which protrude into the benthic boundary layer. For most species of benthic macrofauna the dispersal phase occurs during larval life. Once settled, these animals are sedentary and hence likely to be good indicators of any environmental change which is sufficient to remove macrofauna and replace them with organic enrichment tolerant species. The abundance or density of each taxon is measured per unit area of seabed.

The recognized seasonality of macrofaunal species abundance (e.g. Arntz and Rumohr, 1987) means that special precautions need to be taken to account for this phenomenon, dependant on the type of monitoring goal (Table 1) chosen. We do not recommend to use macrofaunal sampling for the geographical goal of Table 1 to measure the spatial limits of organic enrichment in sediments. This is because the large number of samples required would be prohibitively expensive to analyze taxonomically.

Field sampling

Soft sediment quantitative sampling can be achieved with a wide variety of grabs or corers. A good survey of these devices is presented in Eleftheriou and McIntyre (1984),

although this second edition has become dated. We recommend that grabs of at least $0.1m^{-2}$ be used to sample sediments (Dybern et al. 1976; Wildish 1983; Pohle and Thomas1997). For quantitative sampling any sample where the sediment-water interface is not intact should be discarded and a new sample taken. The grab or corer contents are sieved as soon as possible after collection, preferably on board ship, using pumped seawater to flush away the sedimentary particles. The animals collected on the sieve are preserved in 5-10% buffered formalin in seawater in a sealed bucket. Important information which should be recorded in the field note book and on, and in, the bucket include:

- the date of sampling;
- the relative volume of sediment in the grab/core sample;
- the surface position and depth at the time of sampling from the vessels bridge.

Pohle and Thomas (1997) provide further details of sampling. The use of grab acoustic positioning technology (e.g. McKeown et al. in prep.) allows one to know the precise location where the grab was sampled, and these samples will be of much greater value for archiving (e.g. when acoustic backscatter maps become generally available).

Laboratory analysis

This process involves sorting, identification, counting and weighing. Samples need to be transferred to 70% ethanol or 50% isopropanol to avoid formalin-related damage to calcareous parts used in identification e.g. of bivalves. Sample staining is advantageous in recognizing inconspicuous specimens. Local keys (e.g. Pollock (1998) for the North American east coast) can be used for preliminary identification, but supplemental and more detailed keys must also be available. Any doubtful or "new" species are sent to experts for conformation. Quality assurance procedures must also be implemented (e.g. Pohle and Thomas 1997).The output data consist of a list of species for the whole collection of samples as rows against density or biomass in adjacent columns of locations and/or replicates. Species times density or species times biomass matrices are usefully presented on MS Excel worksheets and need to be linked to pertinent metadata (e.g. position sampled, time, environmental variables).

A number of software packages are available with which to assess the species times density or biomass matrices. We are familiar with PRIMER (Plymouth Routines in Multivariate Ecological Research, available from: 6 Hedingham Gardens, Roborough, Plymouth, PL6 7DX, U.K.). Data can be analyzed using uni- or multi-variate methods. For the site comparison goal (Table 1) of determining whether reference and farm sites have the same macrofaunal distribution and density (null hypothesis), a one-way analysis of variance (ANOVA) can be used. To test for differences between various groupings of macrofaunal community samples the multivariate ANISOM routine can be used. Further details of the methods are given in the user tutorial and in Clarke (1993) and Clarke and Warwick (1994).

Thresholds for management purposes

The responses of the sedimentary environment to high inputs of particulate wastes has been investigated in detail with respect to the pulp and paper industry. Reviews concerning these effects were presented by Pearson and Rosenberg (1978). Despite the obvious differences in type of particulate wastes from pulp mill and fish farm, and in particular the higher rate of biodegradability of fish farm wastes, the environmental responses were similar. Temporal and spatial gradients of response have been described with respect to the benthic communities near point source pulp and paper mill effluents (Pearson and Rosenberg 1978) and fish farms (Gowen and Bradbury 1987; Ritz et al. 1989; Tsutsumi 1990; Johannssen et al. 1994; Wu 1995; Lu and Wu 1998; Brooks & Mahnken 2003a).

The generalized organic enrichment model of Pearson and Rosenberg (1978) is a continuous gradient of successional stages, although it can be divided into four convenient stages of benthic community response (where organic enrichment increases down the page):

Stage	Macofaunal benthic communities
Normal	- stable 'normal' community which is characteristic of the location
Oxic	- transitional between the ecotone point and the normal community
Hypoxic	- peak of opportunists of few species and high density, and the ecotone point where eveness diversity is high and density is lower
Anoxic	- few to no macrofauna, although Pearson and Rosenberg (1978) describe this as azoic

Wildish et al. (1999, 2001) proposed that the stages along the organic enrichment gradient could be used for establishing thresholds for habitat management purposes. One difficulty is that the species of macrofauna are local in distribution, e.g. the opportunists of the hypoxic successional stage differ between the west and east coasts (Table 2). Also an experienced ecologist or taxonomist is generally needed to define the successional stage of a given sample of benthic macrofauna. Although this method clearly demonstrates whether, or not, an environmental change has occurred, a potential difficulty is that it is a high cost method due mainly to the long time required to process samples in the laboratory. Wildish et al. (2001) showed that for two sites in the Bay of Fundy 22 geochemical (redox potential and total sulfide) samples could be processed in the same time as one macrofauna sample.

One approach which could reduce the high cost of macrofaunal sampling is to identify the macrofauna at a higher taxonomic level than genus and species as examined by Karakassis and Hatziyami (2000) in the fish farm context. A concomitant result of this approach is to prevent the use of the organic enrichment model of Pearson and Rosenberg (1978) being applied. A simpler method would be to assess the area of *Beggiatoa* sp. coverage and assess the density of capitellid worms, which would indicate at least the most impacted sediments (equivalent to hypoxic and anoxic zones).

Geochemistry: Total Sulfide in Surficial Sediments

Two electrochemical methods suitable for field deployment have been widely used to indicate the dominance of sulfate reducing bacteria within sediments. Measurements of oxidation-reduction potentials (Eh) in porewater with platinum/reference electrodes (Whitfield 1971) have been widely used for this purpose. A negative Eh indicates anoxia and a positive Eh oxic sediment. Bivariate pairs of Eh/total sulfide data from farm and reference sediments generally demonstrate an inverse relationship when total sulfide, plotted on the x-axis is expressed as $\log_{10}$ and Eh,mV on the y-axis as an arithmetic scale (Hargrave et al. 1997). Recent results have shown that redox probes used for routine monitoring in oxic and anoxic sediments can become "poisoned" and unstable if used again in oxic sediments (Wildish et al. 2004b). This limits their use in defining organic enrichment stages (see below).

An electrochemical method to measure total sulfide in pore water was introduced by Berner (1963). Total sulfide measurements made in the field using this method showed that they were adequately reproducible (e.g. Adams et al. 1972; Wildish et al. 2004b). Adams et al. (1972) also showed that the electrochemical method was consistent with the standard, colorimetric method for total sulfides described by Cline (1969).

Field sampling

Any field sampling device (grab, corer), taken remotely from a support vessel or by SCUBA diver, can be used. The only requirement is that each sample consists of an intact and undisturbed sediment-water interface from which an interfacial sub-sample can be taken. Sub-samples of sediment are withdrawn in a cut-off plastic syringe, so as to exclude air, from within the 0 – 2 cm depth surficial layer. Surficial sampling is thought to be best for monitoring purposes because this is where the most recent impacts on the sediment are likely to be found. Unless analyzed for total sulfides immediately the sub-samples must be "capped" to prevent air entry and immediately stored in an ice chest or refrigerator (Wildish et al. 2004b). Reference localities should be selected following the preliminary survey of benthic habitats.

Laboratory analysis

Refrigerated sub-samples may be stored for up to 48h before analysis. Details of the electrochemical method by ion analysis are given in Wildish et al. (1999; 2004). During analysis the volumetric proportions of sulfide anti oxidant buffer (SAOB): standard or sample, alter the mV response recorded. In order to maintain direct comparability with historic data and among contemporary investigators we suggest that the SAOB:standard (sample) ratio remain at 1:1. Because sulfide standards oxidize on exposure to air frequent checks of the standard solution must be made with an independent method to determine that sulfide levels have not changed.

Thresholds for management purposes

Wildish et al. (2001) proposed geochemical criteria which corresponded to the organic enrichment gradient groupings defined by the benthic macrofaunal communities of Pearson and Rosenberg (1978) resulting from pulp and paper mill pollution. These criteria fit available data from the Bay of Fundy industry and were:

- Normal <300μM $S^{=}$
- Oxic 300 – 1300 μM $S^{=}$
- Hypoxic 1300 – 6000 μM $S^{=}$
- Anoxic >6000 μM $S^{=}$

The geochemical groupings obtained in this classification imply that the macrofauna within them will be of the same successional stage (although the actual species present will reflect the local nature of the species composition, i.e. will be different on west and east coasts). Whether the geochemical criteria, developed on the east coast, can be applied more widely, e.g. on the west coast has been considered by Brooks and Mahnken (2003a). They described an inverse relationship between the number of macro-infaunal taxa found near farms on $\log_{10}$ total sulfide concentration at the same farm, which explained about 50% of the variation. This supports the idea that the organic enrichment gradient is continuous and hence that thresholds can be chosen arbitrarily, although are affected by other unknown factors which contribute to the high variance in the regression. Two other points of difference with organic enrichment gradient thresholds between east and west are in the total sulfide limits for increased abundances of opportunist taxa (see Fig. 9 in Brooks and Mahnken 2003a), which are ~150 – 6000 μM (versus 1300 - 6000μM in the Bay of Fundy), and in the fact that a few tolerant macrofauna do occur where $S^{=}$ >6000μM, yet this is proposed as the azoic zone for macrofauna by Pearson and Rosenberg (1978). These findings suggest that other taxa (e.g. microflora or meiofauna) be sought to define this stage of organic enrichment. They may also suggest that benthic patchiness is confounding the results and/or that there are real differences between west and east coast organic enrichment gradients. Natural levels of total sulfides may vary seasonally (Marvin-DiPasquale and Capone,1998), although this did not appear to be the case in a far-field, mariculture impacted locality, where the interfacial sediment sulfide ranged up to 2500 μM (Wildish et al. 2002).

Geochemistry: Zinc and Copper in Soft Sediments

Observations of trace elements such as zinc (Zn) and copper (Cu) that might be used as specific tracers of waste products released from farms have been measured in water, sediment trap material and surface sediments as an indication of the distance of far-field dispersion of particles released from finfish aquaculture sites (Lewis and Metaxas 1991; Brooks 2001; Sutherland et al. 2002; Brooks and Mahnken 2003b; Yeats 2002; Yeats at al. 2005). Total concentrations have usually been used to determine gradients over distance around farm sites. Without measures of speciation (for example complexation with organic matter) there is no indication of bioavailability by the measurements. There are difficulties in determining threshold levels of metals that lead to sublethal effects of sensitive life history stages of benthic fauna. Despite these limitations, recent observations by Brooks (2001) and Brooks and Mahnken (2003b) in the Broughton area of British Columbia indicate that levels of these metals in surface sediment around farm sites correspond to measured organic enrichment gradients (based on sediment total organic matter, free sulfides and changes in benthic macrofauna communities). Trace elements such as Zn and Cu (and perhaps others) thus have potential to provide additional spatial information to interpret changes in organic matter accumulation around finfish farm sites.

Zinc is commonly added as a nutritional supplement to feed, and copper is used by the industry for antifouling. The difference in sources and some recent field observations indicate that Zn may be useful as a tracer of particulate matter transport while Cu can be used as a tracer of the release of soluble substances such as dissolved nutrients from

farm sites. Elevated concentrations of several other metals including molybdenum (Mo), cadmium (Cd) and uranium (U) have been observed in association with Zn and Cu maxima in sediment cores under farm sites (Smith et al. 2005). There are no clear associations of these metals with direct discharges from aquaculture activities, but elevated concentrations are related to organic enrichment (Cd) and anoxia (Mo and U) in sediments. Many factors affect variability in trace metal concentrations in sediments controlling both bioavailability and toxicity. Metal-sulfide complexes with low solubility are formed in the absence of oxygen (Cooper and Morse 1998; Chapman et al. 1998). Sediment grain size and mineralogy are also fundamental properties determining trace metal concentrations in sediments. Since concentrations increase with decreasing grain size, some means of normalization is required to allow comparison of values in different samples. Lithium (Li), which varies inversely with inorganic grain size, can be used to standardize trace metal levels with respect to grain size to allow small differences in trace metal concentrations to be detected (Yeats 2002; Yeats et al. 2005).

Field sampling

Grabs or cores used for geochemical sampling for trace metal analyses must be chosen to avoid contamination and collect sediment layers either from the surface or from known depths in dated cores. Disturbed samples with mixtures of deeper and surface sediments will alter trace metal concentrations in the uppermost layers. In some cases it may be valuable to obtain a chronology of changes in concentrations over time using stratified coring where the age of sediment with increasing depth is known (Smith et al. 2005). Reference locations are chosen following the preliminary benthic habitat survey of the area.

Laboratory analyses

Inductively coupled plasma mass spectroscopy (ICPMS) is the recommended method for trace metal analysis. Commercial laboratories at reasonable cost can carry out high quality analyses and a single analysis gives results for Zn, Cu, Li, Cd, Mo, U and a number of other elements. Either total acid digestion or strong acid leaching of samples are adequate as long as Li measurements for grain size normalization are made on the same samples using the same digestion techniques (Yeats et al. 2005).

Thresholds for management purposes

Identification of areas where concentrations of contaminants from aquaculture activities exceed natural background concentrations is one potentially useful threshold for management. For sediments, concentrations above background (natural) levels must be quantified with respect to Li in the same sample. Empirical regressions between Li, Zn and Cu must be determined for each area where the method is applied (Yeats et al. 2005). In a relatively large dataset from SWNB (n=120) representative of a range of inshore-offshore sedimentary conditions elevated Zn concentrations could be detected at a 95% confidence level when concentrations were >12 mg kg^{-1} above values expected from empirical regressions. Yeats et al. (2005) concluded that material containing 10% by weight of farm-derived material could be detected if waste material had a Zn content of 200 mg kg^{-1}.

Sutherland et al. (2002) observed variability and asymmetrical footprints for Zn in sediments under and away from net-pens in the Broughton Archipelago that has implications for monitoring for regulatory purposes and habitat management decisions.

Highest values were not always directly under pens, and the extent and shape of the enrichment zone was different from that observed for organic matter. Resuspension, horizontal dispersion, variable sedimentary conditions affecting organic matter accumulation and trace metal diagenesis are factors that could account for the lack of spatial congruity. This emphasizes the problem of basing siting and/or regulatory decisions solely on geochemical observations in sediments at a few locations under netpens or within lease boundaries. Sampling is required over a scale larger than a farm to account for possible redistribution of wastes.

Toxicity is another potentially useful threshold for management action. However, as the bioavailability of metals to macrofauna is dependant on the sulfide levels in sediments (Lee and Lee, 2005) toxic thresholds will be variable.Various national and provincial (or state) agencies have established water and sediment contaminant criteria for protection of aquatic life. In Canada the main compilation of these guidelines is the Canadian Environmental Quality Guidelines established by the Canadian Council of Ministers of the Environment (CCME).

Data for trace metals in sediments under some salmon pens in SWNB and the Broughton Archipelago showed relatively high values (Zn 100 to >400 mg kg^{-1}, Cu 30 to >100 mg kg^{-1}), similar for Zn to those in salmon feed pellets (Brooks and Mahnken 2003b; Chou et al. 2002 and 2004). Concentrations of Zn and Cu under net-pens are usually much higher than those in sediments some distance away (50 to 100 m). But even for maximum levels, in only a few cases did values exceed thresholds identified by the CCME (Anon 1999) as probable effects levels: Zn = 271mg kg^{-1} and Cu = 108 mg kg^{-1} (Yeats et al. 2005).

Photograph and Video Imagery

Still photographs and video recordings can be used to provide 1) a reconnaissance survey of the benthic environment to characterize existing habitats to implement the appropriate substrate-specific monitoring equipment (Fig. 1); 2) calibration information for acoustic surveys where applicable (Clarke et al. 1996); and 3) a quantitative survey to estimate observed waste material, epifaunal abundance, and/or epiflora cover across mixed substrates (Crawford et al. 2001). While geochemical and biological monitoring methods should be applied in predominantly soft-substrates, mixed substrate environments consisting of mosaic patches of soft-sediment, shell hash, boulder fields, and rocky inclines will ultimately rely heavily on video imagery to characterize benthic habitats and determine the potential for organic enrichment. This section of the report will focus on Option 3 which provides a simplistic and cost-effective approach to providing a quantitative assessment of a highly patchy environment that might require a complex and consequently expensive monitoring program according to the monitoring decision tree (Fig. 1). Since the detection of organic enrichment associated with aquaculture activities will be influenced largely by quantitative analytical methods including geochemical triggers (sulfides) and tracers (Zn/Li), it is important that the video monitoring is combined with geochemical monitoring where possible. Management thresholds cannot be derived from changes in observed waste material or biomass alone, but will arise from correlations between epifauna/epiflora abundance and geochemical analyses.

Field sampling

Table 3 provides a summary of a variety of investigations documenting the use of photograph or video techniques to estimate populations of biota or algae across a variety

of substrates. In addition, Table 4 provides the strengths and weaknesses of these techniques categorized according to imaging technology and deployment strategies. Although the tables can be used in tandem to identify an imaging tool suitable for a specific monitoring environment, a high-resolution, practical, and cost-effective imaging tool can be implemented to provide a standard set of analyses for various substrate types.

When considering the quality of imaging technologies, rapid advancements in this field have greatly improved the resolution of underwater images supporting the potential for detailed analyses of the benthos. However, the integrity of the images produced by cutting-edge video equipment can be influenced by both environmental conditions and deployment strategies, thereby hampering analytical efforts and overall data integrity. For example, small-scale ROVs have been towed along transect lines instead of being remotely operated due to drag from high current environments that would prevent an ROV from staying on track and maintaining a constant focal length (Service and Magorrian 1997; Starmans et al. 1999). In addition, more detail was observed from still photos taken from a camera positioned at a constant height above the seafloor relative to those of Hi-8 mm video recordings during a benthic impact assessment study (Collie et al. 2000). Attaching cameras to ROVs or towed video equipment can serve as a backup for data collection as long as the still photographs are taken at a stationary position and fixed focal length during survey transects (Engel and Kvitek 1998). Real-time video coverage of survey transects viewed from the boat deck will help the operator maintain a constant height (focal length) above the seafloor in strong currents or under a grading substrate. In summary, drop cameras (McDaniel et al. 1977; Amos et al. 1997) appear to be a reliable, practical, and cost-effective means of surveying the seafloor in the interim as it can easily and repeatedly monitor a landmarked or geo-referenced station (Beuchel and Gulliksen 2002) over a wide variety of substrate and relief types. Drop cameras provide a standard imaging product using a trigger weight releasing mechanism that is activated at a set distance above the substrate and minimizes substrate disturbance. Cameras can used in tandem with video or ROV deployments which are currently undergoing rapid development in terms of technology and capabilities (Gordon et al. 2004).

Laboratory analysis

Image analysis should provide quantitative estimates of feed pellets, faecal matter, sediment colour, microbial cover, inclusive of *Beggiatoa* sp, epifloral cover, and epifaunal abundance (Crawford et al. 2001). Infaunal organisms may be enumerated when an identifiable part of their body is visible (Starmans et al. 1999). Using available software packages (e.g. OPTIMUS), filters or manual digitization techniques can be used on still photographs and frozen video frames to estimate the aerial coverage of various sediment types, microbial films, and/or algal canopies. Video footage can be frozen at specified frames along a transect to target a geo-referenced area or to reduce the analytical component of the study in the case of densely populated sampling sites. It is considered to be important that all data should be properly geo-referenced.

Thresholds for management purposes

Since microbial, algal, and epifaunal assemblages can show high spatial and seasonal variations, it is important that observed changes in these varaibles are linked to changes in organic enrichment and tracers linked to farm-derived waste material and not used as an independent management threshold. In addition, both the diversity and overall abundance of organisms needs to be considered to detect shifts in populations from oxic

to anoxic conditions as certain organisms may replace a diverse benthic population and proliferate in anoxic, sulfide-rich environments (e.g. *Capitella* sp.).

Benthic Boundary Layer Seawater Sampling (Dissolved Oxygen)

Water column sampling has been an important component of environmental sampling over the past decade under the Modelling-Ongrowing fish farms-Monitoring (MOM) program in Norway (Aure and Stigebrandt 1990; Mäkinen 1991; Wallin and Håkanson 1991; Ervik et al. 1997; Hansen et al. 2001; Nordvarg and Håkanson 2002). This is the sampling method used to assess the status of dissolved nutrient enrichment. However, when hard substrates prevent use of grabs or corers to obtain sediments samples, water from near-bottom within the benthic boundary layer (BBL) may be collected to provide an alternative set of measurements for assessing benthic habitat conditions. We have chosen dissolved oxygen (DO) as the best variable in seawater to indicate organic enrichment effects, since it is important to all aerobic organisms in the near-shore environment.

Field sampling

The locations chosen for sampling should include reference stations in the far-field as well as others close to the fish farm. Samples should be taken from within the benthic boundary layer with a messenger released water bottle or with a moored DO sensor. Care must be taken to avoid introducing air into the samples. The modified Winkler method of Levy et al. (1977) uses a custom-made stopper with a glass extension tube that prevents trapping of air. Semi-continuous electronic recording systems (e.g. a Seacat SBE16) utilizing polarographic electrodes, can also be deployed from moored buoys at the required depth.

Laboratory analyses

Seawater samples for Winkler titration are fixed in the field with manganous and alkaline iodide reagents and analyzed as soon as possible (see in Levy et al. 1977). Hargrave et al. (1993) used polarographic electrodes in temperature controlled injection chambers for small volumes of seawater. Results from the Seacat SBE16 can be downloaded to a laptop. However, it is required to frequently change or clean the polarographic probe to obtain accurate data.

Thresholds for management purposes

Local and far-field factors affecting changes in dissolved oxygen and inorganic nitrogen concentrations in coastal waters and areas with finfish aquaculture development are dealt with in other working papers in this series. Sufficient background data are available for these variables in NF, SWNB and BC both near and distant from aquaculture farm sites (Page et al. 2005; Strain and Hargrave 2005; Stucchi et al. 2002 and Tlusty et al. 2005) that a comparative sampling approach could be used to determine the level of enrichment at a specific farm site. Local dissolved oxygen and inorganic nitrogen levels can be compared with background (far-field) concentrations to assess eutophication effects. For example, in surface waters of the outer Bay of Fundy, as in many temperate coastal areas, dissolved inorganic nitrogen is almost completely removed (concentrations <1 µM) during late summer.

The available reference data for DO in Canada shows that there are natural cycles controlled by physical oceanographic processes, e.g. upwelling, seasonality due to biological factors, tidal and climatic changes (Stucchi et al. 2002; Page et al. 2005). Thus in the Bay of Fundy during the spring/summer bloom, seawater is supersaturated with DO, but in the fall it becomes undersaturated due to the preponderance of heterotrophic metabolism. The Canadian Environmental Guidelines (Anon 1999) suggests a baseline of either 8.0 mgl^{-1} or the lowest naturally occurring level; and that anthropogenic sources should not decrease this level by more than 10%. This pragmatic approach could be adopted for environmental control of the finfish mariculture industry, perhaps with conversion of DO values to a percentage of the DO equilibrium value at the same salinity, temperature and pressure.

FAR-FIELD ENVIRONMENTAL MONITORING METHODS

Benthic Macrofauna of Soft Sediments

Because of the prohibitive cost of processing the large number of grab samples needed in spatial surveys we do not recommend benthic macrofauna to define the geographical limits of organic enrichment impacts. However, the temporal goal (#3 in Table 1) may be feasible. The aim in temporal sampling is to assess the before/after status of sampling locations where, in the intervening period, a fish farm has been constructed and its day to day operations initiated. Whilst not establishing the spatial extent of an impact, this approach may provide a means of monitoring far-field effects if the locations to be sampled are chosen judiciously. As far as we are aware only two studies have led to the recognition of far-field effects involving changes of the benthic macrofaunal community structure which are attributable to salmon culture wastes (Pohle et al. 2001; Wildish and Pohle 2005).

Field sampling and laboratory analysis

The details of sample taking, preservation, sorting, identification and enumeration are as described for near-field sampling. Because the temporal goal is different than for near-field sampling, the strategy employed will also differ. Two approaches are possible:

- Contemporary before/after study. Sampling locations are chosen with the aid of other available information, including sediment type, current patterns, wind activity etc, which helps indicate where fish farm wastes might be transported. Ideally the timing of sampling is just before farm start-up and at annual anniversaries of the first sampling, to avoid seasonal effects.
- Historical before/after study. In this case the sampling locations may have been taken for other purposes, prior to fish farming. Subsequent to fish farm start-up samplings are repeated to give a temporal view of benthic community between the two (or more) periods. Suitable reference locations should be included, which are not impacted by mariculture, to indicate the natural changes occurring in the local area.

Some difficulties experienced with temporal sampling are that it is necessary to assume inter-annual stability (i.e. a stable, equilibrium macrofaunal community) and seasonal effects in species times abundance matrices which are sampled in different seasons must be considered.

Multivariate methods are appropriate for the analysis of macrofaunal and environmental data based on the first approach, i.e. with variables and methods which are comparable between the null and alternative hypothesis tested. These methods involve fewer assumptions when compared to univariate ones (Clarke and Warwick 1994) and allow a greater level of sensitivity to be used in the analysis.

For the second approach recent advances in analytical methods have rendered macrofaunal data from multiple sources more amenable to use where the goal is a temporal one as described above. With univariate methods it is now possible to include data sets collected by different investigators, with different sampling methods, levels of sampling effort and at different times. The more robust methods that have been developed can be applied in environmental impact assessment and in comparing biodiversity (Feral et al. 2003). The new methods (taxonomic distinctness measures, Warwick and Clarke 2001) are based on phylogenetic relatedeness, taking into account the important factor of taxonomic spread of the species. The earlier univariate metrics did not account for taxonomic relatedness, so that species from the same genus would be considered as biodiverse as those species from different families, orders, classes etc; thus missing a major component of biodiversity. The new metrics include average taxonomic distinctness (AvTD) which measures the degree to which species in an assemblage are related to each other (Clarke and Warwick 1998), and variation in taxonomic distinctness (VarTD) which measures the degree to which taxa are over or under-represented in samples (Clarke and Warwick 2001a). Using a master taxonomic list for an appropriate geographic area (e.g. for the Bay of Fundy at: http://gmbis.marinebiodiversity.ca/BayOfFundy/search.html and in draft form for the northwest Atlantic, http://www.huntsmanmarine.ca/narms.htm), where levels up to phylum are included, it is possible to derive an "expected" level of taxonomic distinctness (as mean and confidence limits). Using observed and simulated data from the master list it is possible to test for departure from the expected level of taxonomic distinctness. The statistical framework in the newer versions of Primer allows for both AvTD and VarTD to be combined, thus producing a 2-dimensional plot and fitted simulation envelope, to which real data sets can be compared (Fig.2 A and B). Generally it is found that taxonomic distinctness is reduced at environmentally degraded locations compared to more pristine ones (Warwick and Clarke 1998).

Thresholds for management purposes

Because little work has been done to date we can offer no threshold levels for far-field effects utilizing benthic macrofauna. Further analytical work may confirm practical indicators, e.g. a change in the proportion of molluscs in the macrofaunal sample as a result of organic enrichment (Brooks and Mahnken 2003a). However, likely effects are expected to be subtle (Pohle et al. 2001) and may not be readily discernible by benthic community changes of the organic enrichment gradient. In the case of L'Etang Inlet, before/after historical changes in the far-field involved the disappearance of a mixed community dominated by suspension feeders to one dominated by a diverse assemblage of deposit feeders (Wildish and Pohle 2005). This was consistent with the increased amounts of organic matter produced from fish farms and settling from the water column (Milligan and Law 2005).

Intertidal/Subtidal Macrophytes

Trace metals and inorganic nutrients are readily assimilated by microalgae on surface intertidal sediments and by attached macrophytes (Campbell 2001). Internal reserves accumulated when dissolved nutrients are abundant are used to support growth during times of the year when concentrations are low (Chapman and Craigie 1977). Chopin and Yarrish (1999) and Chopin et al. (2000) claim that intertidal macrophytes can represent an important sink for nitrogen retention in coastal areas. Use of *Laminaria* has been proposed as a method for reducing nutrient release from finfish farms (Subandar et al. 1993). The view is supported by Worm and Lotze (2000) and Worm et al. (2000) who postulated that nutrient cycling and energy flow through macrophytes was a critical process in coastal food web community function. To the extent that nutrient storage by macrophytes (both intertidal and subtidal) buffers against seasonal variations in nutrient availability, these communities may serve as ecosystem bio-monitors of eutrophication. Note however, that nutrient accumulation is dependant on environmental conditions. For example, subtidal macrophytes do not accumulate nutrients in nutrient-replete environments (Anderson et al. 1981; Gagné et al. 1982).

Robinson et al. (2005) in their study of increased macroalgal cover, due to eutrophication from aquaculture, which smothered clams, *Mya arenaria*, in SWNB sampled two intertidal areas proximate to salmon farm sites. Enhanced Zn:Li ratio tracers were found in surface sediments from the depositional intertidal area where *Ulva* sp., cover had increased, but no enrichment was found at the second location where sediment grain size was coarser. The association of Zn with fine-grained deposits suggests transport to the intertidal zone by a fine sediment fraction as would be expected from results of depositional models. The observations imply that if particulate Zn is transported from farm sites over distances >1 km, then dissolved inorganic nutrients would also reach the intertidal zone. Relatively high levels of dissolved nitrogen in inshore vs. offshore areas in SWNB are discussed in another working paper in this series (Strain 2005).

Field sampling and laboratory analysis

Water, sediment and macrophytes from intertidal areas may be collected without remote sampling equipment. However, time series or spatially distributed sampling is necessary to provide a synoptic view of macrophyte cover. This can be achieved by aerial photographic surveys (Robinson et al. 2005). Variability in measurements at single locations or over a limited time period may be difficult to interpret or attribute to a specific aquaculture source. Macrophytes can be collected and stored frozen until analyses for elemental composition and trace metals following acid digestion as described for sediments. Various sample types should be collected from as many different intertidal zones (high, mid and low) as possible to ensure some degree of spatial resolution. This will also provide data from a range of sediment types. Sediment grain size usually varies from more coarse-grained deposits in lower intertidal levels to more fine-grained sediments in upper regions. SPM and sediment samples can be analyzed for trace metals using ICPMS with concentrations normalized for Li as described for subtidal sediments. Concentrations of inorganic nutrients and trace metals in macrophyte tissues should be reported on an ash-free weight basis.

Thresholds for management purposes

As with recommendations for interpreting variables collected in the BBL, no universally applicable thresholds are available and a comparative approach based either on before/after or reference/treatment samples for evaluating the differences in macrophyte cover or biomass observed in the intertidal zone is proposed. Robinson et al. (2005) showed that the proportion of the intertidal area occupied by *Enteromorpha* sp dramatically increased as a result of nutrient enrichment effects. Even with sampling restricted to two intertidal areas, Robinson et al. (2005) were able to interpret results of trace metal analysis to infer that factors responsible for enrichment originated from a salmon farm >1 km away.

Towards Habitat Management Using Acoustic Methods

Recent advances in acoustic technology, largely developed by geologists, make it possible to describe surficial sediments at geographically meaningful scales relatively cheaply. It is highly likely that these techniques will also be of value in benthic biology to describe habitats/communities/seascapes which will be key in managing the marine environment. We cannot describe here exactly how to use these methods because they are still under development. Instead we present a brief review of the current developments.

Improvements in resolution of single beam (SB) sonar allowed attainment of 100% coverage with swath techniques such as side scan (SS) and multibeam (MB) sonars. Resolution with the latter is no longer dependant on the spacing of single points or tracks but rather on instrument and operational specifics (i.e. beam angle, depth, tow speed, refresh rate, positioning accuracy).

Modern sonar tools range from simple "fish finders" to the highly sophisticated and more expensive multibeam systems. Data treatment and interpretation also varies from visual inspection of analogue or digital output to sophisticated signal decomposition and analysis. The resulting output can be further analyzed using various clustering techniques and/or entered into a GIS or other spatial analysis software for display or further analysis.

Sonar signals provide direct information about the topography of the substrate and its depth. Since substrate type and complexity affect the strength and shape of the return echo (backscatter) information on seafloor characteristics can also be obtained from the sonar signal. The simplest characterization used in low end echo sounders or obtained from visual inspection of backscatter intensity, provides an indication of hard (rock) versus soft (silt, sand) bottom with an intermediate category of mixed (rubble, cobble) substrates. A simple classification scheme such as this is adequate to characterize substrates as depositional or erosional for evaluation of locations of potential accumulation of aquaculture wastes or to determine grab sample locations.

Methods for extracting or inferring additional information from acoustic signals have been developed to provide more detailed classification of substrates and to relate them to benthic community structure. These efforts at habitat classification using acoustics have met with varying degrees of success. In general, they are suitable for application to gently sloped substrates with no highly three dimensional features (Freitas et al. 2003; Brown et al. 2004). Most require extensive groundtruthing, careful instrument calibration and a high degree of operator expertise.

A brief overview of the tools, methodologies and means of substrate characterization using acoustic methods is presented below with the advantages and disadvantages of each. All of these approaches have been tested at aquaculture sites. However, there have been no standardized applications of these methodologies to aquaculture environmental assessment or environmental effects monitoring. The selection of a specific method will depend on several factors including scale, heterogeneity of the area of interest, cost, availability of equipment and degree of sophistication of post-acquisition signal processing that is required.

Low end depth sounder (fish finder)

A simple, inexpensive protocol for seafloor mapping and substrate classification has been developed for coral reefs in Malaysia (Kvernevik et al. 2002) using an inexpensive sounder, a GPS and towed video camera. The methodology provides an inexpensive procedure for obtaining and mapping good quality data of bottom type and topography applicable to shallow coastal areas. Resolution is determined by the sonar footprint (a function of beam angle and depth) and thus varies with depth. Kvernevik et al. (2002) also provide tips to optimize resolution, sample density and sampling and video processing time. This protocol is now routinely used to monitor coral reef habitat in Malaysian parks (Kvernevik et al. 2002) and is readily adaptable to aquaculture lease sites.

A similar approach has been developed to map depth and substrate types and to guide grab sampling at aquaculture sites in Newfoundland (Barkhouse et al. in prep.). In this work a GPS and fish finder were combined to collect depth and position data, while the "white line" (backscatter strength indicator) was used to classify the substrate into 3 categories (rock, mixed/rubble, soft). Ground-truthing with grab samples and underwater video demonstrated the accuracy of the classification system. Data collection was rapid and a 20 ha lease site could be mapped in half a day. Grab sample time was also significantly reduced in comparison to the currently applied grid sampling pattern since depositional areas could be targeted. In this instance, some manual data acquisition were used, although the protocol could easily be automated by the method of Kvernevik et al. (2002).

Single beam sounder with signal interpretation

Acoustic surveys with automated signal interpretation are usually carried out with higher end sounders. Some systems are portable whereas others are platform specific and require vessel specific calibration. Costs thus range from moderate to high depending on equipment and platform requirements. In recent years, a number of approaches to using backscatter signals to classify substrate and habitat have been developed. The best known (and most tested) of these are RoxAnn™ and QTC View™. Both approaches use statistical techniques to extract and analyze information for the return signal. Once the return signal has been characterized (using 2 elements – RoxAnn™, or 3 principal components – QTC View™) the resulting data matrix can be classified into empirically determined habitat types (RoxAnn™) or statistically and then related to empirically determined habitat or substrate types (QTC Impact™). While both approaches are useful for characterizing large areas of the sea floor, each suffers from some disadvantages and both are adversely affected by the potential for mismatch between scale of habitat complexity and scale of sample collection (track spacing).

RoxAnn™ substrate classification has been used to support indications of limited sedimentation near certain aquaculture sites in the Mediterranean (MacDougal and Black 1999). However Wilding et al. (2003) have found that RoxAnn™ may not be suitable for monitoring change over time, since in Scottish coastal waters repeat sampling did not always produce the same substrate maps possibly due to unpredictable influence of vessel speed on the output.

QTC View™ and QTC Impact have been used to classify coastal substrates around the world (Anderson 2002; Ellington 2002), although Morrison et al. (2001) found that acoustically detected transition zones did not always correspond to visual observation of the seafloor. They suggest that either the substrate acoustic characteristics do not match the visual characteristics of interest, or there is a mismatch of scale between the features of interest and the acoustic samples. Freitas et al. (2003) found that QTC View™ classified fine grained sediment well and can be related to benthic community structure, but the system performed less well for coarse grained sediments. Recently, Hewitt et al. (2004) found that the proprietary software QTC Impact™ used to classify substrate types did not match biological classifications as well as alternative methods (Q- Package) proposed by Legendre et al. (2002) and available as freeware from the internet. These findings have sparked a lively debate in the literature (Preston and Kirlin 2003; Legendre 2003) about the most suitable approach to acoustic signal classification. Hewitt et al. (2004) also suggest that while the rules for groundtruthing remotely sensed terrestrial data are well developed, the situation for acoustic sensing of aquatic habitat is complicated by the influence of depth on resolution and the lack of direct links between the acoustic signal and community characteristics. Foster-Smith et al. (2004) compared RoxAnn™ and QTC View™ at two sites in the English Channel and found that while both systems give similar output, RoxAnn™ consistently gave slightly better levels of performance than QTC View™. They concluded that careful data processing is critical to avoid artifacts and that this approach is good for large scale mapping but less useful for fine scales because of the limits of single beam resolution (Foster-Smith et al. 2004).

Swath bathymetry

Sidescan sonar produces a swath image of the seafloor. Tracks can be positioned to overlap providing 100% coverage. Backscatter information can be analyzed visually or using greyscale intensity. The resulting information can be classified and related to substrate characteristics if desired. Side scan sonar equipment, both portable and vessel-mounted, is available at moderate cost, and the size of the survey area covered will be platform specific to a certain extent. Signal processing does require a certain expertise. While visual examination of the resulting greyscale backscatter maps can be used to distinguish hard versus soft bottoms and substrate relief, additional substrate classification requires more sophisticated post acquisition processing (Cutter et al. 2003). Hewitt et al. (2004) found that data matching between sidescan and benthic video was relatively easy in contrast to the match with single beam surveys which were more problematic.

Multibeam sonar uses acoustic signals from an array of sources to improve coverage of the seafloor. Overlapping swaths and DGPS positioning result in 100% coverage. Resolution is a function of depth and position precision. The resulting backscatter information can be mapped or analyzed as greyscale intensity (Tlusty et al. 2000a) to determine hard and soft bottoms. As with sidescan, more sophisticated signal processing can provide additional categories for classification. Multibeam surveys of aquaculture sites in Bay d'Espoir, Newfoundland, Southwest New Brunswick and the Broughton

Archipelago, British Columbia have demonstrated that waste accumulation patterns associated with cage operations can be determined dependant on depth, substrate type and bottom relief (Tlusty et al. 2000a; Wildish et al. 2004a; Sutherland et al. in prep.) The relationship between backscatter intensity and percent organic matter may be difficult to quantify or highly site specific. While high resolution substrate characterization can be acquired for large areas in relative short time scales both data acquisition, and signal processing require expensive equipment and significant expertise. Multibeam surveys by the Canadian Hydrographic Service can provide valuable information for aquaculture site selection and environmental assessment; however, the methodology is currently prohibitively expensive and demanding of specialized equipment and expertise for routine site monitoring.

Acoustic signal interpretation of swath bathymetry and backscatter strength is often done visually (Wildish and Fader 1998) or qualitatively (Edwards et al. 2003) however, quantitative techniques are also applied (Tlusty et al. 2000a; Cutter et al. 2003). Usually some form of greyscale analysis is used to characterize backscatter strength. The resulting output can then be empirically related to substrate and/or habitat characteristics (Cutter et al. 2003).

Swath bathymetry may eventually be the most appropriate approach for examining changes over time, and is the only one capable of spatial scale, particularly for environments with heterogeneous relief and substrates. It is difficult to monitor changes in seafloor characteristics over time using maps with < 100 % coverage since interpolation may change unless exactly the same points or transects are sampled each time. This is particularly true for complex topographies and habitats where care must be taken to match survey resolution to the scale of the features of interest and to substrate heterogeneity. Swath bathymetry allows the creation of digital elevation maps (DEM) of the seafloor. Subsequent maps of the same site can be used to detect changes in substrate characteristics or changes in elevation (i.e. due to deposition of wastes). Resolution of the data will determine the magnitude of change that can be detected. This can range from 0.1 m with careful use of an inexpensive sounder (Kvernivik et al. 2002) to 0.08 m for multibeam surveys adjusted to datum points (Stirling and Roy 2000). Care must be taken to use the same bench marks and to correct for tidal height. This approach has been used in coastal Newfoundland to determine the quantity of mine tailings deposited on the seafloor following a tailings dam breach or from direct tailings deposition to the sea. Stirling and Roy (2000) compared before and after DEM's of two hydrographic surveys, the first completed using soundings and the second more recent survey using multibeam data. By subtraction of the early DEM from the more recent one, they were able to obtain a Differences Digital Elevation Model (DDEM) that could be integrated to determine the volume of tailings in the marine environment.

CONCLUSIONS

The strengths and weaknesses of near-field monitoring methods are summarized in Table 5. For soft sediments three methods are proposed: macrofauna and two geochemical methods. All of these methods are scientifically defensible and lend themselves to statistical testing. All three methods have threshold levels associated with them, although heavy metals have a single concentration threshold for Cu and Zn, which is perhaps less useful because it may exclude subtle effects across the organic enrichment gradient which can affect sedimentary functioning. Total sulfide measurements are more cost effective in defining the organic enrichment gradient, but lack the co-incident information (e.g. trophic

types) contained in macrofaunal data. As far as we are aware the relative cost of the heavy metal determination in sediments has not been estimated.

U/W photography and videography are the only benthic methods available for mixed and hard substrates. Yet these methods are not well known and developed scientifically and cannot at the present time be linked to thresholds of effect. Nor to our knowledge have comparative costs been estimated. For benthic boundary layer seawater sampling, which may be considered as a possible alternative method where hard substrates predominate, we consider that DO threshold levels are the most practical for habitat management purposes. Measurement methods are well established, scientifically defensible, and capable of being used to choose between null and alternative hypotheses. Threshold effect levels for DO are variable with temperature and salinity and hence season and location and this deters use of a universal DO threshold. Adoption of the Canadian Environmental Quality Guidelines (Anon1999) threshold of not more than 10% below the lowest natural level of DO, dictates that background data be available to establish the local "natural levels" as is the case in the Bay of Fundy (Page et al. 2005).

The studies which we have reviewed to detect far-field effects of organic enrichment from mariculture are among the first to establish this link, and consequently there are no established thresholds and cost estimate comparisons. We believe that it is preferable to treat every study as a new research project, since the full range of effects in the far-field environment have not yet been identified.

Finally, we recommend that a two tier level of environmental monitoring be applied, as is done in many other countries, e.g. in the Norwegian monitoring program mentioned earlier in this report. This will allow a relatively simple preliminary monitoring of all farms (using U/W videography, total sulfides and *Beggiatoa* sp. cover), then a subsequently more detailed field survey (including macrofauna, zinc and copper and dissolved oxygen) at those locations where the preliminary work suggests that an environmental problem may be, or is, occurring.

FURTHER RESEARCH

We emphasize that research in environmental monitoring is an active and ongoing process and that new findings and possible changes to current recommendations should be re-considered annually. Listed below, in no particular order of merit, are areas where we believe that further research is required.

1. Maps of sediment acoustic properties are required in areas where marine finfish aquaculture is currently practiced or likely to develop in Canada to provide synoptic assessments of the proportion of hard and soft bottom substrates. The information can be used for modelling the proportion of farm-derived particulate wastes deposited locally or far-field.
2. New methods are required to quantify processes of resuspension that redistribute fine material produced locally by finfish aquaculture sites over larger areas.
3. Continued development of photographic, video and sediment profile imaging techniques is required with ground-truthing in areas where new finfish aquaculture development is likely to occur.
4. Geochemical methods using Li normalized measurements of Zn and Cu in SPM and surface sediments need to be applied in different intertidal areas to confirm that the

method provides a general technique to detect the proportion of farm-derived wastes in SPM and sediment samples.

5. The Bay of Fundy mariculture industry has proposed to expand fish culture to deeper, more offshore sites. Research is required to determine the major environmental risks to other users of the marine environment here, and how they can be monitored and mitigated.
6. The promising new bioindicator tools mentioned in the text (taxonomic distinctness) are being assessed in Britain (R. Clarke pers. comm.) for use in a regulatory context. Taxonomic distinctness methods should also be tested and validated in Canada with respect to the aquaculture industry.
7. What new methods can be devised to monitor macrofauna on hard substrates? This should include methods to assess crevice macrofauna and the use of artificial surfaces (panels) as a way to detect organic enrichment effects.
8. Because of the conflicting results mentioned in the text regarding macrofaunal differences and sulfide levels which separate the oxic and hypoxic stages of the gradient, between east and west coasts, we propose further studies to identify if a universal threshold of sulfide (between 150 to 1300 µM) exists.
9. New, cost effective, methods should be developed to measure the geographic limits of organic enrichment effects in sediments. One possible method using acoustic backscatter contrast has been proposed (Wildish et al. 2004a), although it would probably be limited to soft, depositional sediments.
10. A standardized procedure for U/W videography and framework for describing threshold limits for organic enrichment effects should be developed. The comparative cost effectiveness of the method should also be determined.
11. Are size measurements of meiofauna, as presented by Duplisea and Hargrave (1996), a more cost effective way of monitoring organic enrichment than taxonomic diagnosis of macrofauna? One result of using size measurements of meiofauna or macrofauna would be that there was no established relationship available with the organic enrichment gradient.

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Table 1. Monitoring goals used in detecting and measuring organic enrichment effects resulting from mariculture.

Goals	Effect measured	Hypothesis
1. Practical	Determines a relative impact	None, it triggers remediation or other management activity if a specified threshold (reference point) for a variable is exceeded
2. Site comparison	Difference between treatment/reference sites	H_0 reference = treatment site H_1 reference ≠ treatment site
3. Temporal	Before/after status	H_0 reference = treatment at t_0 H_1 reference ≠ treatment at t_1
4. Geographical	Limits of impact	H_0 reference condition throughout the study area H_1 reference and impacted area delimited within the study area

Table 2. Characteristic benthic macrofaunal opportunistic species tolerant of organic enrichment under and near salmon farms.

British Columbia		New Brunswick	
Species	**Reference**	**Species**	**Reference**
Nebalia puggetensis	Brooks & Mahnken (2003a)	*Capitella capitata*	Hargrave et al. (1995)
Capitella capitata		*Nuculana tenuisulata*	
Sigambra tentaculata		*Nephtys neotena*	
Schistomeringos sp.			
Aoroides sp.		*Nucula proxima*	Pohle et al. (2001)
Pseudotanais oculatus		*Nucula delphinodonta*	

Table 3. A summary of photograph and video studies categorized according to deployment strategy and technology application.

Technique	Water depth	Substrate type	Biota	Authors
Drop camera		Soft sediment	Substrate	Amos et al. 1997
Drop camera	<250 m	Silt, sand, clay	Epifauna	McDaniel et al. 1977
Drop camera	14 to 45 m	Sand	Epifauna	Piepenburg & Schmid 1997
Drop camera	15 m	Rock	Algae, Epifauna	Gulliksen & Beuchel 2002
Diver-deployed camera	12 m	Canyon rock walls	Sponges, Algae, Inverts	Ayling 1983
Diver-deployed camera	1 m	Sea walls	Oysters, Ascidians	Dalby & Young 1992
Diver-deployed camera	7 m	Pilings	Epifauna	Keough 1984
Diver-deployed camera	20 m	Inclined rock walls	Ascidians	Svane & Lundalv 1982
Diver-deployed video	Up to 15 m	Cobble, sand, silt	Scallops, Epifauna	Barbeau et al. 1996
Diver-deployed video	7.5 m	Cobble, sand, silt	Scallops, Epifauna	Hatcher et al. 1996
Diver-deployed video	5 to 12 m	Rock, boulder, sand	Coral, Algae, Inverts	Harriott et al. 1999
Diver-deployed video	1.5 to 3.0 m	Rock, coral, sand	Coral, Algae, Inverts	Carleton & Done 1995
Diver-deployed ROV-video	Nil	Boulder, sand, mud	Epifauna	Service & MaGorrian 1997
Towed georeferenced - video	Max 50 m	Mud, sand	Epifauna	Strong & Lawton 2004
Towed georeferenced - video	Up to 40 m	Gravel/cobble, kelp	Substrate, Veg, Fauna	Morris & Power 2004
Towed video	GB	Gravel, sand	Scallops, Epifauna	Stokesbury et al. 2004
Towed sledge video	30 to 40 m	Sand, mud	Epifauna	Hughes & Atkinson 1997
Towed video, photographs	40 to 80 m	Gravel, cobble, sand	Epifauna	Collie et al. 2000
Towed video	200 m	Bedrock to silt	Substrate, Epifauna	Gordon et al. 2000
Towed ROV	30 to 370 m	Fine-coarse grained	Epifauna	Starmans et al. 1999
ROV transects, still photos	90 to 180 m	Rocks, sand, silt	Substrate, Epifauna	Engel & Kvitek 1998
ROV transects and Diver-deployed video	>20 m < 20 m	Sand	Pellets, Faeces, Sed, Biofilms, Algae, Fauna	Crawford et al. 2001

Table 4. Characteristics of various imaging techniques with associated costs or strengths and weaknesses
High = ***, Low = *, Strong = ***, Weak = *, 0

Characteristics	Drop camera	Diver-deployed camera	Diver-deployed video	Towed-video	ROV
Cost of equipment purchase	*	*	*	** / ***	** / ***
Cost to deploy equipment	*	* / **	* / **	** / ***	** / ***
Operation and deployment logistics	*	**	**	** / ***	** / ***
Type of vessel support	*	*	*	* / ***	* / ***
Operating conditions: Max depth	> 30 m	< 30 m	< 30 m	> 30 m	> 30 m
Operating conditions: Max current					
Ability to target transects/stations	**	***	***	**	* / ***
Ability to carry out repeated sampling	**	***	***	**	* / ***
Ability to survey a sloping seafloor	**	***	***	** / ***	** / ***
Ability to maintain focal length	***	***	***	**	* / **
Discrete or continuous sampling	Discrete	Discrete	Transects	Transects	Transects
Georeferencing capability	**	**	**	** / ***	** / ***
Entanglement factor	*	**	**	**	***
Benthic disturbance potential (impact on soft-bottom and structural species)	*	*	*	**	**

Table 5. Strengths/Weaknesses of organic enrichment monitoring methods based on sediment sampling and grouped according to applicable goals (Numbers refer to Table1). Strong = ***, Weak = *, 0 = not applicable ? = unknown

Operational criteria	Environmental monitoring method							
	Conventional Macrofauna		Total S$^{=}$		Zinc/ Copper		U/W Photo/Video	
Goals	1-3	4	1-3	4	1-3	4	1-3	4
Soft substrate	***	*	***	*	***	*	**	*
Mixed substrate	0	0	0	0	0	0	**	*
Hard substrate	0	0	0	0	0	0	**	*
Scientific defensibility	***	*	***	*	***	*	**	*
Statistical testability	***	*	***	*	***	*	**	*
Threshold availability	***	*	***	0	**	*	?	?
Cost effectiveness	*	0	***	0	?	0	?	?

Fig. 1. Decision tree for selecting sampling methods based on substrate type (Sutherland, 2004)

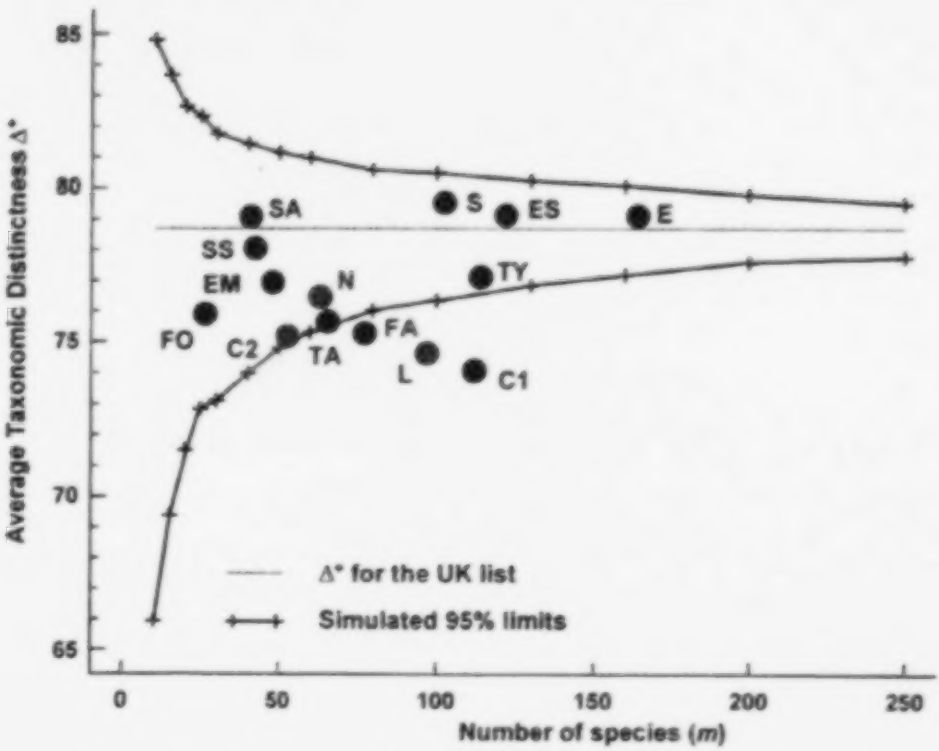

Fig. 2A. Funnel plot with mean and 95% probability limits of simulated taxonomic distinctness values derived from a master species list of UK nematodes. Observed distinctness values are superimposed as black circles, with three samples falling outside the expected norm (from Warwick and Clarke 1998). Note how the mean is largely independent of sample size.

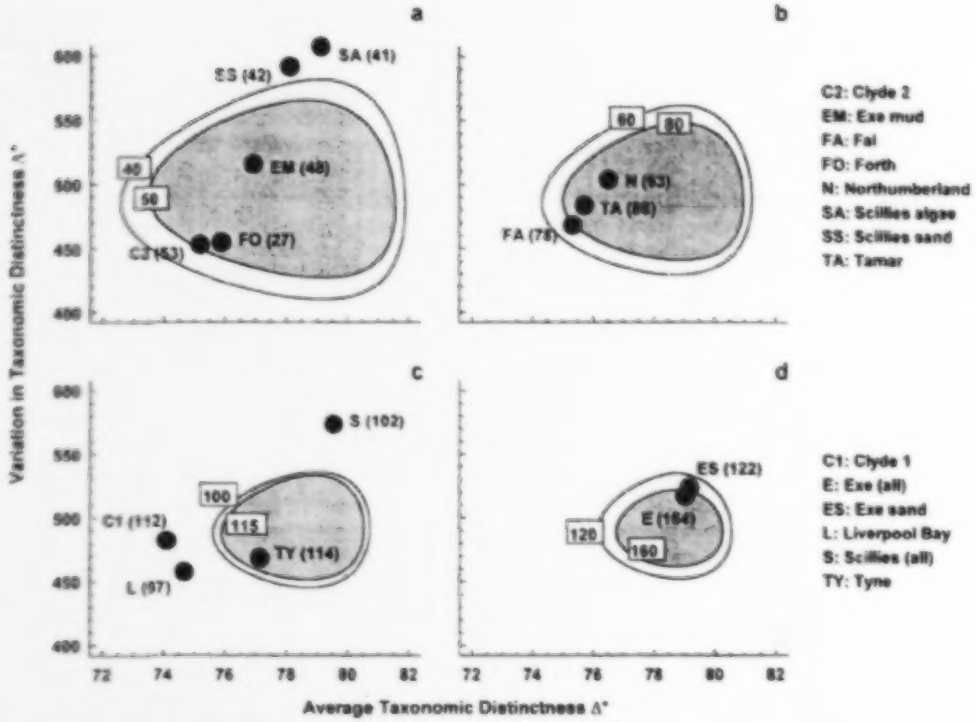

Fig. 2B. Ellipse plots with 95% probability regions based on average taxonomic distinctness and variation in taxonomic distinctness derived from master species list, using different sublist sizes (numbers in rectangles). Superimposed observed distinctness values (black circles) from different locations indicate which samples fall outside the norm (from Clarke and Warwick 2001).

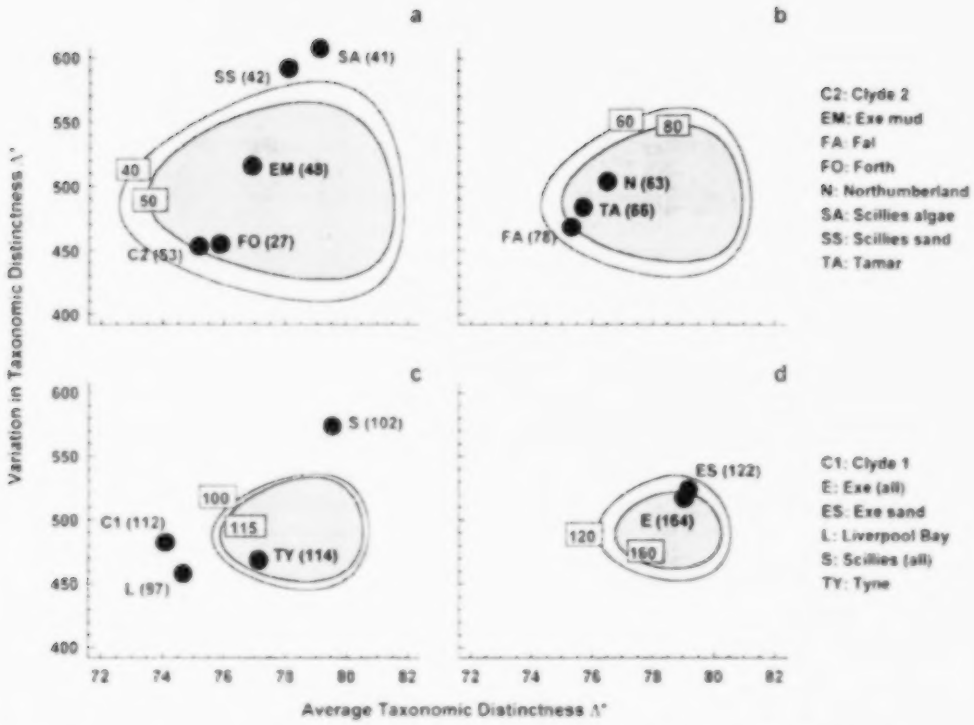

Fig. 2B. Ellipse plots with 95% probability regions based on average taxonomic distinctness and variation in taxonomic distinctness derived from master species list, using different sublist sizes (numbers in rectangles). Superimposed observed distinctness values (black circles) from different locations indicate which samples fall outside the norm (from Clarke and Warwick 2001).